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REMARKS

STATUS OF THE CLAIMS

Claims 1-14 were pending in this application. Claims 13 and 14 have been withdrawn from consideration. Claims 1, 3, and 4 have been amended. Following entry of the amendments claims 1-12 will be pending and at issue.

SUPPORT FOR AMENDMENTS TO THE CLAIMS

Claim 1 is amended to replace the equation $\hat{\text{Var}}(\hat{\mu}) = \hat{\sigma}_\varepsilon^2 + \hat{\mu}^2 e^{\hat{\sigma}_\eta^2}\left(e^{\hat{\sigma}_\eta^2} - 1\right)$, where $\hat{\mu}^2 = (y - \alpha)^2$,

with the following

$$Var\{y\} = \mu^2 e^{\sigma_\eta^2}\left(e^{\sigma_\eta^2} - 1\right) + \sigma_\varepsilon^2$$

wherein μ is the amount of the biological molecule and

$$y = \alpha + \mu e^{\eta} + \varepsilon$$

and wherein each model quantity is replaced by an estimate thereof.

Support for this amendment can be found throughout the specification as filed, e.g., Equation 3 on page 5, line 2; the definition of μ on page 4; and Equation 1 on page 4.

Claim 1 is also amended to insert the language "of a proportional error component, η" in step e. Support for this amendment can be found throughout the specification as filed, e.g., at page 4, line 29.

Claims 3 and 4 are amended to correct inadvertent and/or typographical errors, using the mathematical operator "x" to replace the "x" to clarify that it is a mathematical operation and surrounding that mathematical operation with parentheses.

The amendments to the claims therefore add no new matter and entry is respectfully requested.

ELECTION/RESTRICTION REQUIREMENT

Pursuant to the restriction requirement made final and election of claims 1-12, Applicant withdraws claims 13 and 14 with entry of this amendment. Applicant reserves the right to file subsequent applications claiming the canceled subject matter. In addition, the claim withdrawals

should not be construed as abandonment or agreement with the Examiner's position in the Office Action.

IDS

Applicant notes with appreciation the Examiner's thorough consideration of the references cited in the IDS (Form 1449) submitted on March 7, 2002.

REJECTIONS UNDER 35 U.S.C. § 112, SECOND PARAGRAPH

Claims 1-12 were rejected under 35 U.S.C. § 112, second paragraph as allegedly indefinite for a number of reasons. Applicant addresses these rejections as follows.

Specific to claim 1, step (g), the Examiner stated that "the parameters utilized in the equations have not been defined ... due to the presence of circumflex symbols or cap-like symbols on top of the parameters in the equation." Applicant respectfully disagrees and notes that one of skill in the art readily recognizes that a cap symbol, "^", represents an estimate of the term over which the cap symbol is placed. For example, σ_ε is a model parameter; the same parameter with a cap symbol is an estimate of that parameter. Applicant believes that the claims are not indefinite as originally drafted.

Specific to claim 1, step (g), the Examiner stated that "the equation following the term "where" causes the claims to be not commensurate in scope with the specification...(and) has not been found in the specification." Applicant respectfully disagrees. As described in the specification on page 4, the term "μ" is the expression level in arbitrary units, and points out that one of skill can readily derive the equation following the term "where" in claim 1, step (g)

$$\hat{\mu}^2 = (y - \alpha)^2$$

from, e.g., Equation 1 on page 4:

$$y = \alpha + \mu e^{\eta} + \varepsilon .$$

where an estimate of the expression level, "μ" , is calculated by assigning the value of zero to the error parameters "ε" and "η." These error parameters by definition have average values of zero as is known to one of skill in the art.

However, without agreeing with Examiner's position, but in the interest of furthering prosecution, Applicant has amended claim 1, step (g) to replace

$$\hat{\mathrm{Var}}(\hat{\mu}) = \hat{\sigma}_\varepsilon^2 + \hat{\mu}^2 e^{\hat{\sigma}_\eta^2}\left(e^{\hat{\sigma}_\eta^2} - 1\right), \text{ where } \hat{\mu}^2 = (y - \alpha)^2.$$

with the language

$$\underline{Var\{y\} = \mu^2 e^{\sigma_\eta^2}\left(e^{\sigma_\eta^2} - 1\right) + \sigma_\varepsilon^2}$$

wherein μ is the amount of the biological molecule and

$$\underline{y = \alpha + \mu e^{\eta} + \varepsilon}$$

and wherein each model quantity is replaced by an estimate thereof.

The model equation is model Equation 3, found on page 5 of the specification, and represents the variance of response y at concentration μ (e.g., the amount of biological molecule, e.g., the expression level).

Specific to claims 3 and 4, step c, Examiner stated that "the recitation of "X" causes the claims to be vague and indefinite because it is not clear whether "X" is a variable or a mathematical operation. As "X"" is a mathematical operation, Applicant has amended claims 3 and 4, replacing the lower case ""x" with the mathematical operator "x" and surrounding that operation with parentheses.

Applicant believes that the claims as amended are not indefinite and respectfully requests withdrawal of this rejection.

REJECTIONS UNDER 35 U.S.C. § 112, FIRST PARAGRAPH

Claims 1-12 are rejected under 35 U.S.C § 112, first paragraph, as allegedly containing subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. Applicant respectfully disagrees.

In paragraph 13 of the office action, the Examiner states that

> Claim 1 recites a method for estimating the precision of measurements. However, the instant specification lacks any guidance as to how one of skill in the art to use the estimated precision of measurement generated by the method of the instant invention. It is acknowledged that the instant specification discloses the equation of step (g) and the parameters being used in said equation (pages 13-14); however,

> said specification does not disclose what type of measurements are being represented by the parameters. For example, the specification discloses equation 19 (page 14) as directed to generic measurements and values. One question comes to mind is what measurements or values are being applied to equation 19 for the estimating the precision of measurements. Due to the high level of skill requirement and the lack of guidance how one of skill in the art for determining what type of measurements are to be used with the equation of step (g), one of skill in the art to would not be able to practice the instant invention without undue experimentation.

Applicant's invention is a method to provide estimates of the variance of, e.g., a method for determining the precision of, measured intensity in microarray-derived measurements. The types of data that are analyzed include microarray-derived gene expression experiments. In this type of experiment, a high measured intensity indicates a high level of gene expression, and a low measured intensity indicates a low level of gene expression.

Applicant respectfully points out that the specification is fully enabling for the claimed invention. The specification clearly discloses how one of skill in the art can use the method of the instant invention at page 3, lines 29-34:

> Applications of the model developed in the present invention pertain to detection limits, categorization of genes as expressed or unexpressed, comparison of gene expression under different conditions, sample size calculations, construction of confidence intervals, and transformation of expression data for use in multivariate applications such as classification or clustering.

The types of measurements represented by the parameters are disclosed in the specification at the bottom of page 4:

> ...y is the intensity measurement, μ is the expression level in arbitrary units, α is the mean background (mean intensity of unexpressed genes), η is the proportional error that always exists, but is noticeable at expression levels significantly above zero, and ε represents the additive error that always exists but is noticeable mainly for near-zero expression levels.

The measurement of values that are applied to the equations for estimating the precision of measurements are fully disclosed in the specification. The value of "y" is the measured intensity on a microarray, as described in the preceding quote from the specification. Estimating the standard deviation of low level measurements is described beginning at page 5, line 11:

> The easiest way to estimate the standard deviation σ_ε of the low level measurements is from replicate blanks (negative controls). Data are generated

> using an array identical to the array on which samples will be run, and a blank (comprising components identical to the sample components in all ways except for the presence of sample nucleic acid, which is omitted from the blank) is loaded onto the array, and processed in a manner identical to the procedures used with an actual sample. In some instances, it is possible to use the same array sequentially for obtaining negative control and sample data.

Another method for estimating the standard deviation of low level measurements is described beginning at page 7, line 7:

> If there are no negative controls, σ_ε can be estimated by pooling the variance estimates of genes that have low expression levels. For this, one would use the raw expression values and not the logarithms.

Estimating the mean background is described on page 6, line 4:

> The mean intensity of the negative controls is a suitable estimate of α, the mean background.

Estimating the standard deviation of high level measurements is described beginning at page 6, line 6:

> The parameter σ_η can be likewise estimated from the standard deviation of the logarithm of high level replicated measurements.

Accordingly, Applicant believes that the instant specification clearly provides guidance as to how one of skill in the art is to use the estimated precision of measurement generated by the method of the instant invention (e.g., evaluating the precision of gene expression data) and that the specification discloses what measurements and values are being represented by the parameters of the equations (e.g., the measured intensity and estimates of the expression level, mean background and standard deviations of error). Applicant requests withdrawal of the ground of rejection.

In paragraph 14, the Examiner states that

> Claims 3 and 4, step (c), recite a step for the variable c wherein c is equal or greater than 2, and equal or less then 3. Further, it is acknowledged that the specification discloses the use of the instant invention where in c is 2.5 and 3 (page 13, line 10). However, the specification does not provide any disclosure of how the value of 2.5 or 3 is determined. The instant specification does not provide adequate guidance to one of skill in the art to determine the value of c to practice the claimed invention without undue experimentation.

Applicant respectfully points out that the parameter *c* is not "determined" but rather selected anew with each iteration of the thresholding algorithm. As defined beginning at page 10, line 7 of the specification, the parameter *c* is the number of standard deviations above the mean or median. The parameter *c* is used in a thresholding algorithm to determine the cutoff point. The cutoff point is the expression measurement below which the analyst decides to not analyze data, e.g., expression measurements below the cutoff point are assumed to be unreliable and/or noise specific to the array.

For the purposes of the invention, *c* can be set at 2, 2.5 or 3 for each iteration of the thresholding algorithm. These values for *c* are reasonable given that, e.g., it has been suggested that genes exhibiting at least 3-fold changes in differential expressions in cDNA arrays are deemed significant. Which value of *c* (e.g., 2, 2.5, or 3) is selected by the analyst depends on the desired stringency of the gene expression experiment.

Therefore, Applicant requests withdrawal of this rejection.

CONCLUSION

Withdrawal of the pending rejections and reconsideration of the claims are respectfully requested, and a notice of allowance is earnestly solicited. If the Examiner has any questions concerning this Response, the Examiner is invited to telephone Applicant's representative at (415) 875-2316.

Respectfully submitted,
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